Abstract & Introduction

The FluoroPhenyl stationary phase has long been marketed as a phase that offers alternative, or orthogonal, selectivity to a C18. The FluoroPhenyl phase offers unique selectivity by incorporating strongly electronegative fluorine atoms on a phenyl ring (Figure 1). In addition to the traditional reversed-phase dispersive interactions, this phase also exhibits shape selective, polar, cation-exchange and even HILIC retention mechanisms which aid in selectivity of specific analytes.

In this presentation we aim to demonstrate the useful and alternate retention of the FluoroPhenyl stationary phase. We chose several relevant target analytes which we plan to use to exemplify the unique retention characteristics of the FluoroPhenyl phase when used in either HILIC or reversed-phase mode. All of these analytes have been pursued due to either poor retention, poor resolution, or both on a traditional C18 phase.

Using the Hydrophobic Subtraction Model[1] to Characterize the FluoroPhenyl Stationary Phase

When analyzing the results of the Hydrophobic-Subtraction Model for the FluoroPhenyl, it can be determined that in addition to hydrophobic retention, a FluoroPhenyl phase also exhibits very low steric hindrance. While there is still much to be understood about steric hindrance it appears to be at least one contributing factor to the shape selectivity of a stationary phase. Additionally, the FluoroPhenyl is capable of very strong cation-exchange interactions which allows retention and selectivity of small polar compounds and cationic species.

Figure 2: Hydrophobic-Subtraction Model Values for FluoroPhenyl Stationary Phase on 2.7 µm Raptor™ Silica

<table>
<thead>
<tr>
<th>Column</th>
<th>H</th>
<th>S*</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>C-7</th>
</tr>
</thead>
<tbody>
<tr>
<td>FluoroPhenyl</td>
<td>0.691</td>
<td>-0.114</td>
<td>0.065</td>
<td>-0.058</td>
<td>0.392</td>
<td>1.423</td>
</tr>
</tbody>
</table>

Reversed-Phase Retention of Basic Compounds

Figure 3: Structures of Test Probes Analyzed on Raptor™ FluoroPhenyl Stationary Phase

4-Methylimidazole (4-MEI) is a small polar compound that is used as a caramel coloring in food and beverages. In an acidic solution, 4-MEI has a positive charge, making it very amenable to cation-exchange. When a high proportion of acetonitrile is used in the mobile phase, this molecule displays HILIC-like retention on the FluoroPhenyl (Figure 6). When using electrospray ionization mass spectrometry (ESI-MS), using a high percentage of an organic solvent in the mobile phase eluent increases the efficiency of the desolvation process, increasing sensitivity significantly (Figure 7).

Figure 7: Sensitivity and Retention Comparison of 4-MEI in Reversed-Phase and HILIC Modes on Raptor™ FluoroPhenyl

Conclusions

- Shape selectivity properties of the FluoroPhenyl stationary phase allow for the separation of epimers not easily accomplished on a C18.
- The FluoroPhenyl phase allows retention based on hydrophobicity as well as the cation-exchange mechanism. This results in a mixed-mode column which supports reversed-phase and HILIC analyses.
- By utilizing the cation-exchange mechanism and high organic mobile phases, increased retention and sensitivity may be seen for compounds such as 4-MEI.